Claims

1. A method of identifying a ligand which can mediate the biological activity of a target protein via inhibition of the binding of a target protein to a binding partner which comprises

(a) screening a first combinatorial library comprising a plurality of first member ligands for binding to the target protein, thereby identifying one or more target-binding ligands,

(b) screening a second library comprising a plurality of second member ligands for the ability to inhibit the binding of one or more of said target-binding ligands to said target protein, thereby obtaining one or more inhibitory ligands, where said second library is not identical to said first library, and

(c) determining which of the inhibitory ligands can mediate a biological activity of the target protein,

wherein, when the ligands are peptides, they are not more than 41 amino acids long.

2. The method of claim 1 wherein the first combinatorial library is composed of peptides, peptoids, nucleic acids, or combinations thereof.

3. The method of claim 1 in which the first combinatorial library is composed of peptides, peptoids, or a combination thereof.

4. A method according to claim 1, wherein the first combinatorial library is composed of peptides.

5. The method of claim 4 in which the first library is a biased peptide library, or a combination of two or more different biased peptide libraries, but not an unbiased peptide library.

6. The method of claim 5 which comprises, in step (a), screening a structured panel of biased combinatorial peptide libraries, each library having one and only one constant residues at a first fixed position that is the same for all of the libraries and is, within the middle 50% of the peptide, but the amino acid assigned to said first position is not the same

in all libraries of said panel, and as a result of such library-to-library variation, said panel collectively presents all possible genetically encoded peptides of a predetermined length, and only peptides of that length.

7. The method of claim 6 wherein said peptides are of the form

$$(Xaa)_m\text{-}R1\text{-}(Xaa)_n,$$

where Xaa is any amino acid, R1 is the amino acid at said first fixed position, m and n are each in the range of 2-20, and m and n do not differ by more than two.

8. The method of claim 7 in which each Xaa is, independently, any naturally occurring amino acid.

9. The method of claim 7 in which each Xaa is, independently, any genetically encoded amino acid.

10. The method of claim 5, said structured panel further characterized in that in each library, there are two and only two constant residues,

one such residue being at a first fixed position that is the same for all of the libraries and is within the middle 50% of the peptide,

and the other constant residue is at a second position, but the location of said second position is varied so that said second position scans all residue positions except for said first fixed position, whereby the panel is composed of subpanels, characterized in that within each subpanel said second position is fixed, and

the amino acid assigned to said first position is not the same in all libraries of said panel, and the amino acid assigned to said second position is not the same in all libraries of a given subpanel.

11. The method of claim 10 where as a result of such library-to-library variation, said panel collectively presents all possible genetically encoded peptides of a predetermined length, or all possible genetically encoded peptides of a predetermined length which do not contain cysteine, and only

peptides of that length.

12. The method of claim 5 which comprises, in step (a) screening a structured panel of biased combinatorial peptide libraries, consisting of a plurality of biased combinatorial linear peptide libraries, each library comprising a plurality of different peptides, all peptides of said panel being of the same length, there being one and only position in said peptides which is both (1) for each library, the same amino acid (a "constant" amino acid) in all peptides of that library, and (b) not the same amino acid in all libraries of said panel, said position being fixed for all peptides in all libraries of said panel, wherein said fixed position is (a) at least five residues from both ends of the peptides or (b) within the middle 50% of the peptides,

each library being a separate and physically distinct entity from all other libraries of the panel, in which the peptides are displayed on viruses.

13. The method of claim 12, wherein there is no position which is the same amino acid for all peptides of the panel.

14. The method of claim 12 wherein said peptides are of the form

$$L_1\text{-}(Xaa)_m\text{-}R1\text{-}(Xaa)_n\text{-}L_2,$$

where L_1 and L_2 are each independently chosen from the group consisting of nothing and a subsequence of one or more amino acids, said subsequence being the same for all peptides of the panel,

R1 is the amino acid at said first position, and

m and n are independently chosen from the range of 2 to 20.

15. The method of claim 12 wherein if L_1 or L_2 is a subsequence of one or more amino acids, the subsequence is not more than three amino acids.

16. The method of claim 15 in which L_1 is nothing, SS-, or SR-, and L_2 is nothing or -SR.

17. The method of claim 5 which comprises, in step (a), screening a structured panel of biased combinatorial linear peptide libraries, each library comprising a plurality of different peptides, all peptides of said panel being the same length, each library having at least two constant residue

positions, one at a first position and the other at a second position,

where the first position is fixed for all libraries in the panel, and is assigned the same residue for all peptides in any given library, but libraries of the panel collectively present a plurality of different residues at said first position,

where said first position is (a) at least five amino acids from both ends of the peptides, or (b) is in the middle 50% of the peptides,

where said panel comprises a plurality of subpanels, each comprising a plurality of libraries, and in each subpanel, the location of the second position is constant, but said location varies from subpanel to subpanel so the second positions of said subpanels, collectively scan a plurality of residue positions other than said first position,

where the second position is assigned the same residue for all peptides in a given library but the libraries of a given subpanel collectively present a plurality of different residues at said second position,

where if said libraries comprise more than two constant residue positions, the constant residue positions other than said first and second positions are constant for all peptides in said panel,

where one or more of the other positions of said libraries are variable positions, at which a given library exhibits a plurality of different residues as a result of sequence variation from peptide to peptide,

each library being a separate and physically distinct entity from the other libraries of the panel.

18. The method of claim 17 in which the second positions collectively scan all residue positions which are variable across the panel as a whole except for said first position.

19. The method of claim 5 which comprises, in step (a), screening a structured panel of biased combinatorial linear peptide libraries, each library comprising a plurality of different peptides, all peptides of said panel being the same length, each library having at least two biased residue positions, one at a first position and another at a second

position, the amino acids allowed in each library at said biased positions being only a subset of the set of amino acids allowed at the remaining positions of said library, and also being only a subset of the set of amino acids allowed at that biased position in the panel as a whole,

where the first position is fixed for all libraries in the panel,

where said first position is (a) at least five amino acids from both ends of the peptides, or (b) is in the middle 50% of the peptides,

where said panel comprises a plurality of subpanels, each comprising a plurality of libraries, and in each subpanel, the location of the second position is constant, but said location varies from subpanel to subpanel so the second positions of said subpanels collectively scan a plurality of residue positions other than said first position,

where if said libraries comprise more than two constant residue positions, the constant residue positions other than said first and second positions are constant for all peptides in said panel,

each library being a separate and physically distinct entity from the other libraries of the panel.

20. The method of claim 19 in which the second positions collectively scan all residue positions which are variable across the panel as a whole except for said first position.

21. The method of claim 5 which comprises, in step (a), screening a structured panel of biased combinatorial linear peptide libraries, each library comprising a plurality of different peptides, all peptides of said panel being the same length, each library having at least two biased residue positions, one at a first position and another at a second position, the amino acids allowed in each library at said biased positions being only a subset of the set amino acids allowed at the remaining positions of said library, and also being only a subset of the set of amino acids allowed at that biased position in the panel as a whole,

where the first position is fixed for all libraries in the panel,

where said first position is (a) at least five amino acids from both ends of the peptides, or (b) is in the middle 50% of the peptides,

where each library is obtained by mixing a plurality of different mixed oligonucleotides, each oligonucleotide comprising one fully variable codon and one less variable codon, the position of the less variable codon varying so that said plurality collectively scan also positions other than said first fixed position, said less variable codon encoding the second position of each peptide,

where if said libraries comprise more than two constant residue positions, the constant residue positions other than said first and second positions are constant for all peptides in said panel,

each library being a separate and physically distinct entity from the other libraries of the panel.

22. The method of claim 21 in which the second positions collectively scan all residue positions which are variable across the panel as a whole except for said first position.

23. The method of claim 5 which comprises, in step (a), screening a structured panel of biased combinatorial peptide libraries, said panel consisting of a plurality of biased combinatorial linear peptide libraries, each library comprising a plurality of different peptides, all peptides of said panel being of the same length, there being in said peptides, either one position which is or two positions which are (1) for each library, the same amino acid (a "constant" amino acid) in that position in all peptides of that library, and (2) not the same amino acid in that position in all libraries of said panel,

at least one of said positions being fixed for all peptides in all libraries of said panel, said fixed position being (a) at least five residues from both ends of the peptides or (b) within the middle 50% of the peptides,

each library being a separate and physically distinct entity from all other libraries of the panel,

in which the peptides are displayed on viruses.

24. The method of claim 19 where the panel has overall diversity which is the same at each position of the peptides,

and a given library has a diversity at a biased position which does not exceed 3.

25. The method of claim 21 in which, in a given library, the first and second positions are amino acids belonging to one and only one of the following groups:

1	W	TGG
2	F, Y	T(T,A)T
3	P	CCT
4	H	CAT
5	D, E	GAX
6	K, R	A(G,A)A
7	N	AAT
8	Q	CAA
9	L, I, V	(G,A,C)TT
10	M	ATG
11	S, T	A(G,C)T
12	A, G	G(G,C)T
13	C	TGT

26. The method of claim 25 in which, in a given library, the first fixed position belongs to one and only one of groups 1-7 and 13.

27. The method of claim 12 wherein the overall diversity of the panel at the fixed position of the peptides is the same as the overall diversity of the panel at each of the other positions of the peptides.

28. The method of claim 5 which comprises, in step (a), screening a structured panel of biased combinatorial peptide libraries, each library having one or two constant residues, wherein, in each component library, at a first fixed position within the middle 50% of the peptide, the amino acid assigned to said first position is constant within said component library, is not the same in all libraries of said panel, and as a result of such library-to-library variation, said panel collectively presents all possible genetically encoded peptides of a predetermined length, and in which the peptides are displayed on viruses.

29. The method of claim 26 wherein said peptides are of the form

$(Xaa)_m$-R1-$(Xaa)_n$,

where Xaa is any amino acid, R1 is the amino acid at said first

fixed position, and m and n are each in the range of 2-20.

30. The method of claim 28, said structured panel further characterized in that in each library, a second position is held constant, but the location of said second position is varied so that said second position scans all residue position except for said first position, whereby the panel is composed of subpanels in which said first and second positions are fixed, and where, in each subpanel, the amino acid assigned to said second position is constant within said component library, but varies from library to library within said subpanel.

31. The method of claim 5 which comprises, in step (a), screening a biased combinatorial peptide library of the form

$(Xaa)_m$-Cys,

where m is greater than or equal to 5, and Xaa is any amino acid.

32. The method of claim 1 in which the first combinatorial library comprises nucleic acids.

33. The method of claim 1, wherein the first library has a diversity of at least 10^3 different sequences.

34. The method of claim 1, wherein the first library has a diversity of 10^3 to 10^9 sequences.

35. The method of claim 1 in which the first library has a diversity of at least about 10^6 different sequences.

36. The method of claim 1 in which the target-binding ligands obtained in step (a) are tested in a suitable biological system for the ability to interact with the target protein so as to mediate its biological activity and only the effective ligands are used in screening step (b).

37. The method of claim 1 in which the inhibitory ligands obtained in step (b) are tested to determine whether their inhibitory action is attributable to their binding the target protein or to their binding the target-binding ligand.

38. The method of claim 1 in which the step (a) ligands which bind the target are separated or otherwise distinguished from ligands which do not bind the target.

39. The method of claim 1, further comprising screening the second library, before, during or simultaneously with step (b) for the ability of the second member ligands to bind to the

target protein.

40. The method of claim 1 in which the second member ligands are screened for the ability to competitively inhibit the binding of said target-binding first member ligands to the target protein.

41. The method of claim 1 wherein the second library is screened for the ability to inhibit the binding of a plurality of target-binding first member ligands of said first combinatorial library.

42. The method of claim 1 wherein the second library is screened for the ability to inhibit the binding of a substantially all of the target-binding first member ligands of said first combinatorial library.

43. The method of claim 41 where, in step (b), the target protein is contacted with a mixture comprising said plurality of target-binding first member ligands.

44. The method of claim 1 where the first library is obtained by expression in cells of genes encoding expression products, each expression product comprising a first member ligand.

45. The method of claim 44 in which the first member ligands are displayed on the surface of said cells or on the outer coats of phage produced by said cells.

46. The method of claim 1 where the first library is obtained nonbiologically.

47. The method of claim 1 where the first library is screened in solution phase.

48. The method of claim 1 where the first library is screened in solid phase.

49. The method of claim 48 in which the first member ligands are immobilized on solid nonliving surfaces.

50. The method of claim 1 in which the target protein is one associated with human cytomegalovirus.

51. The method of claim 50 in which the target protein is the DNA polymerase accessory protein UL44.

52. The method of claim 51 in which at least one of the first member ligands used in step (b) is a peptide comprising the sequence

(E/D/N/Q)-(H/R/K)-(V/L/I/M)-C-(S/T/A/G)-W-G-W-G-(R/K/H)-C.

53. The method of claim 50 in which at least one of the first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:19-30.

54. The method of claim 1 in which the target protein is human MDM2.

55. The method of claim 54 in which at least one of the first member ligands used in step (b) is a peptide comprising the consensus sequence

F-X-D-X-W-X-X-L

where X is any amino acid.

56. The method of claim 55 in which at least one of the first member ligands used in step (b) is a peptide comprising the sequence S-F-T-D-Y-W-R-D-L-E-Q or a conservative mutant thereof.

57. The method of claim 54 in which at least one of the first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:43-46 and 50-53.

58. The method of claim 1 in which the target protein is an enzyme.

59. The method of claim 58 in which the target protein is a protein kinase.

60. The method of claim 59 in which the protein kinase is human protein kinase c beta II.

61. The method of claim 60 in which at least one of the first member ligands used in step (b) is a peptide comprising at least one of the consensus sequences

W-Phi-C-Pho-G-X-F/L-C and

W-T-C-V/I-N-C

where X is any amino acid, "Phi" is a hydrophilic amino acid and "Pho" is a hydrophobic amino acid.

62. The method of claim 60 in which at least one of the first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:32-42.

63. The method of claim 58 wherein the target protein is a transferase.

64. The method of claim 58 in which the target is an isomerase.

65. The method of claim 58 in which the target protein is a synthetase.

66. The method of claim 58 in which the target protein is a transfer RNA synthetase.

67. The method of claim 66 in which the target protein is ProRS.

68. The method of claim 67 in which at least one of the first member ligands used in step (b) is a peptide comprising the sequence

S-R-W-C-acid-Pho-W-P-Phi-X-X-G-C-S-R (SEQ ID NO:62)

where X is any amino acid, "acid" is an acidic amino acid, "Pho" is a hydrophobic amino acid, and "Phi" is a hydrophilic amino acid.

69. The method of claim 67 in which at least one of the first member ligands used in step (b) is a peptide comprising the sequence S-R-(D/E)-W-G-F-W.

70. The method of claim 67 in which at least one of the first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:54-61, 64 and 66.

71. The method of claim 66 in which the target is TyrRS.

72. The method of claim 71 in which at least one of the first member ligands used in step (b) is a peptide comprising the sequence YXWP (SEQ ID NO:93) or YWWPDWG (SEQ ID NO:94), where X is any amino acid.

73. The method of claim 71 in which at least one of the first member ligands used in step (b) is a peptide comprising at least one sequence selected from the group consisting of

(1) Y-Phi-W-P-W

(2) Y-Phi-W-P-Phi

(3) (Y/F)-(S/T/G/A/H)-W-P-(W/G/D/S/P) and

(4) (Y/F/W/L)-W-W-P-(D/E/S/N)-W-G.

74. The method of claim 71 in which, at least one of the

first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:67-92.

75. The method of claim 58 in which the target is beta-glucosidase.

76. The method of claim 75 in which, at least one of the first member ligands used in step (b) is a peptide comprising the sequence PWP.

77. The method of claim 75 in which at least one of the first member ligands used in step (b) is a peptide comprising the sequence

P-W-P-(I/V)-Y.

78. The method of claim 75 in which, at least one of the first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:95-104.

79. The method of claim 58 in which the target is carboxypeptidase.

80. The method of claim 79 in which at least one of the first member ligands used in step (b) is a peptide comprising the sequence

P-G-W-W.

81. The method of claim 79 in which, at least one of the first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:105-107.

82. The method of claim 58 in which the target is alcohol dehydrogenase.

83. The method of claim 82 in which, at least one of the first member ligands used in step (b) is a peptide comprising a sequence selected from the group consisting of SEQ ID NOs:108-113.

84. The method of claim 67 in which the target is biotynylated ProRS.

85. The method of claim 84, in which at least one of the first member ligands used in step (b) is a peptide comprises a sequence selected from the group consisting of SEQ ID NOs:114-117.

86. The method of claim 1 in which the target protein is a transmembrane receptor.

87. The method of claim 1 in which the target protein is a nuclear receptor.

88. The method of claim 87 in which the nuclear receptor is an estrogen receptor.

89. The method of claim 88 in which at least one of the peptides of said first library comprises the sequence LXXLL.

90. The method of claim 88 in which at least one of the first member ligands used in step (b) is a peptide comprising at least one sequence selected from the group consisting of

(1) W-Pho-R-L-Phi-D-Pho-P-W-G

(2) C-F-F-W-D and

(3) L-X-X-L-L

where Pho is a hydrophobic amino acid, Phi is a hydrophilic amino acid, and X is any amino acid.

91. The method of claim 88 in which at least one of the first member ligands used in step (b) is a peptide comprising at least one sequence recited in Tables 11-1, 11-2 or 11-3.

92. The method of claim 87 in which the nuclear receptor is a glucocorticoid receptor.

93. The method of claim 87 in which the nuclear receptor is a testosterone receptor.

94. The method of claim 87 in which the nuclear receptor is a retinoic acid receptor.

95. The method of claim 87 in which the target protein is an androgen receptor.

96. The method of claim 87 in which the target protein is a progestin receptor.

97. The method of claim 87 in which the target protein is a steroid receptor.

98. The method of claim 87 in which the target protein is a thyroid receptor.

99. The method of claim 87 in which the target protein is a vitamin D receptor.

100. The method of claim 87 in which the nuclear receptor is homodimeric.

101. The method of claim 87 in which the nuclear receptor

is heterodimeric.

102. The method of claim 87 in which the nuclear receptor is estradiol-activated.

103. The method of claim 4 in which the target is a nuclear receptor.

104. The method of claim 4 in which the target is an enzyme.

105. The method of claim 1 wherein the second library has a diversity of at least 100 different structures.

106. The method of claim 102 wherein the second library has a diversity of at least 11,200 different structures.

107. The method of claim 1 wherein at least one ligand of said second library has a molecular weight which is less than that of $(Gly)_{11}$.

108. The method of claim 1 wherein at least one ligand of said second library has a molecular weight which is less than that of $(Gly)_5$.

109. The method of claim 1 wherein every ligand of said second library has a molecular weight which is less than that of $(Gly)_{11}$.

110. The method of claim 1 wherein every ligand of said second library has a molecular weight which is less than that of $(Gly)_5$.

111. The method of claim 1 wherein the average molecular weight of the ligands of the second library is less than the average molecular weight of the ligands of the first library.

112. The method of claim 1 wherein the second molecular weights of the ligands of the library are all less than those of the ligands of the first library.

113. The method of claim 1 in which the second library comprises peptoids.

114. The method of claim 1 wherein the second library is not biopolymeric.

115. The method of claim 1 in which the second library comprises members which are not selected from the group consisting of peptides, peptoids and nucleic acids.

116. The method of claim 1 in which the second library is composed of members which are not peptides.

117. The method of claim 1 in which the second library is composed of members which are not peptides, not peptoids, and not nucleic acids.

118. The method of claim 1 in which the second library is composed of nonoligomeric compounds.

119. The method of claim 1 in which the second library is a combinatorial library.

120. The method of claim 1 in which the second library is a heterocyclic combinatorial library.

121. The method of claim 120 in which the second library comprises cyclic compounds containing one heteroatom.

122. The method of claim 121 in which the heteroatom is a heteronitrogen.

123. The method of claim 122 in which heteronitrogen compounds are selected from the group consisting of pyrroles, pyrrolidines, pyrrolines, prolines, indoles, beta-carbolines, pyridines, isoquinolines, quinolones, beta-lactams, and azabicyclo [4.3.0] nonen-8-one amino acids.

124. The method of claim 121 in which the heteroatom is a heterooxygen.

125. The method of claim 124 in which the heterooxygen compounds are selected from the group consisting of furans, pyrans and gamma-butyrolactones.

126. The method of claim 121 in which the heteroatom is a heterosulfur.

127. The method of claim 126 in which the heterosulfur compounds are sulfolenes.

128. The method of claim 120 in which the second library comprises compounds with two or more heteroatoms.

129. The method of claim 128 in which the multiple heteroatom compounds comprises two or more heteronitrogens.

130. The method of claim 129 in which the multiple heteronitrogen compounds are selected from the group consisting of imidazoles, pyrazoles, piperazines, benzodiazepines, 1,4-benzodiazepine-2,5-diones, hydantoins, dihydropyrimidines, 1,3-disubstituted-5,6-dihydropyrimidine-2,4-diones, cyclic ureas, cyclic thio ureas, quinazolines, triazoles, and purines.

131. The method of claim 128 in which the multiple

heteroatom compounds comprises compound with both at least one heteronitrogen and at least one heterooxygen.

132. The method of claim 130 wherein the second library comprises compounds selected from the group consisting of diketomorpholines, isoxazoles and isoxazolines.

133. The method of claim 128 in which the compounds comprise compounds with both at least one heteronitrogen and at least one heterosulfur.

134. The method of claim 133 in which the compounds comprise compounds selected from the group consisting of thiazolidines, dihydrothiazoles, 4-melathiazanones, and benzisothiazolones.

135. The method of claim 117 in which the second library comprises is a benzodiazepines.

136. The method of claim 1 in which the second library is a simple combinatorial library.

137. The method of claims 1 in which the second library is not a combinatorial library.

138. The method of claim 1 in which the first combinatorial library has a greater diversity than the second library.

139. A method of identifying a ligand which can mediate the biological activity of a target protein via inhibition of the binding of a target protein to a binding partner which comprises

(a) screening a first combinatorial library comprising a plurality of first member ligands for binding to the target protein, thereby identifying one or more target-binding ligands,

(b) screening a second library comprising a plurality of second member ligands for the ability to inhibit the binding of one or more of said target-binding ligands to said target protein, thereby obtaining one or more inhibitory ligands, and

(c) determining which of the inhibitory ligands can mediate a biological activity of the target protein

wherein, when the first member ligands are all peptides, step (a) comprises screening a structural panel of biased combinatorial peptide libraries.

140. A method of identifying a ligand which can mediate the biological activity of a target protein via inhibition of the binding of a target protein to a binding partner which comprises

(a) screening a first combinatorial library comprising a plurality of first member ligands for binding to the target protein, thereby identifying one or more target-binding ligands,

(b) screening a second library comprising a plurality of second member ligands for the ability to inhibit the binding of one or more of said target-binding ligands to said target protein, thereby obtaining one or more inhibitory ligands, and

(c) determining which of the inhibitory ligands can mediate a biological activity of the target protein

wherein, when the first member ligands are all peptides, the first library is an unbiased library, or is biased at not more than two amino acid positions.

141. A method of identifying a ligand which can mediate the biological activity of a target protein via inhibition of the binding of a target protein to a binding partner which comprises

(a) screening a first combinatorial library comprising a plurality of first member ligands for binding to the target protein, thereby identifying one or more target-binding ligands,

(b) screening a second library comprising a plurality of second member ligands for the ability to inhibit the binding of one or more of said target-binding ligands to said target protein, thereby obtaining one or more inhibitory ligands, and

(c) determining which of the inhibitory ligands can mediate a biological activity of the target protein

wherein, when the first member ligands are all peptides, at least some of those peptides do not contain internal cysteine residues.

142. A method of identifying a ligand which can mediate the biological activity of a target protein via inhibition of the binding of a target protein to a binding partner which comprises

(a) screening a first combinatorial library comprising a plurality of first member ligands for binding to the target protein, thereby identifying one or more target-binding ligands,

(b) screening a second library comprising a plurality of second member ligands for the ability to inhibit the binding of one or more of said target-binding ligands to said target protein, thereby obtaining one or more inhibitory ligands, and

(c) determining which of the inhibitory ligands can mediate a biological activity of the target protein

wherein, when the ligands are peptides, they do not comprise antibody-like domains.

143. The method of claim 142 in which the second library comprises ligands which are not peptides.

144. The method of claim 142 in which the first combinatorial library comprises ligands selected from the group consisting of peptides, peptoids and nucleic acids, and the second library comprises ligands which are not selected from that group.

145. A method of identifying a pair of ligands, the first ligand of said pair binding to a target protein, and the second ligand of said pair inhibiting the binding of the first ligand to said target protein, which comprises

(a) screening a first combinatorial library comprising a plurality of first member ligands for binding to the target protein, thereby identifying one or more target-binding ligands,

(b) screening a second library comprising a plurality of second member ligands for the ability to inhibit the binding of one or more of said target-binding ligands to said target protein, thereby obtaining one or more inhibitory ligands,

where said second library is not identical to said first library

where the first ligand of the pair is one of said target binding first member ligands of said first library, and

where the second ligand of the pair is one of the second member ligands of said second library,

wherein, when the ligands are peptides, they are not more than

41 amino acids long.

146. A method of identifying a pair of ligands, the first ligand of said pair binding to a target protein, and the second ligand of said pair inhibiting the binding of the first ligand to said target protein, which comprises

(a) screening a first combinatorial library comprising a plurality of first member ligands for binding to the target protein, thereby identifying one or more target-binding ligands,

(b) screening a second library comprising a plurality of second member ligands for the ability to inhibit the binding of one or more of said target-binding ligands to said target protein, thereby obtaining one or more inhibitory ligands,
where said second library is not identical to said first library
where the first ligand of the pair is one of said target binding first member ligands of said first library, and
where the second ligand of the pair is one of the second member ligands of said second library,
wherein, when the ligands are peptides, they do not comprise antibody-like domains.